



Transfer and long-term persistence of plasmids encoding antimicrobial resistance in wastewater treatment plant microbial communities

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Publication date:
2018

Document Version
Peer reviewed version

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Citation (APA):

Li, L., Dechesne, A., Madsen, J. S., Nesme, J., Kalckar, A., Sørensen, S. J., & Smets, B. F. (2018). *Transfer and long-term persistence of plasmids encoding antimicrobial resistance in wastewater treatment plant microbial communities*. Abstract from 17th International Symposium on Microbial Ecology, Leipzig, Germany.

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Title: Transfer and long-term persistence of plasmids encoding antimicrobial resistance in wastewater treatment plant microbial communities

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There is increasing concern that wastewater treatment plants (WWTPs) may contribute to the dissemination of antimicrobial resistance genes (ARGs). Indeed, there transfer of plasmid-encoded ARGs might occur as human intestinal and environmental bacteria mix. Assuming that most enteric bacteria die off, ARG survival depends on transfer to and maintenance in WWTP adapted bacteria. Here, we explore, at the community level, the notion that plasmids cannot maximize both their within-population maintenance and their transfer proficiency, but that a tradeoff exists between these two properties.

We provide the first assessment of the transfer and maintenance of typical ARG-carrying plasmids in a WWTP activated sludge microbial community. This community was challenged with *Escherichia coli* carrying one of three GFP-tagged plasmids spanning multiple important incompatibility groups: pKJK5 (IncP), R27 (IncH) and R64 (IncI). Transconjugants were separated by fluorescence-activated cell sorting and identified by 16S rRNA gene amplicon sequencing. pKJK5 transferred to a broad phylogenetic range of bacteria, spanning 13 phyla, while transfer of R27 and R64 was limited to *Enterobacteriaceae*. Following initial transfer, the communities were serially propagated over at least 60 generations, and plasmid persistence monitored. For pKJK5, high plasmid-carrying fractions (up to 10%) were transiently observed, which decreased and remained above 1% by the end of the experiment. In contrast, R27 and R64 never reached high incidence. The description of the diversity of the plasmid carriers along the serial propagation experiment is pending, but will provide a unique insight into the dynamics of contrasting plasmids in complex microbial communities.